UNITED STATES ENVIRONMENTAL PROTECTION AGENCY
WASHINGTON, DC 20460

OFFICE OF PREVENTION,
PESTICIDES AND TOXIC
SUBSTANCES

MEMORANDUM

DATE: December 13, 2011

SUBJECT: Efficacy Review for Benefect Botanical Daily Cleaner Disinfectant Spray;
EPA Reg. No. 84683-3;
DP Barcode: D393313

FROM: Thao Pham
Product Science Branch
Antimicrobials Division (7510P)

THRU: Dr. Tajah Blackburn, Team Leader
Product Science Branch
Antimicrobials Division (7510P) 12/15/11

TO: Jacqueline Campbell-McFarlane PM34/Jaclyn Carl
Regulatory Management Branch II
Antimicrobials Division (7510P)

APPLICANT: OhSo Clean, Inc.
315 Pacific Avenue
San Francisco, CA 94111

FORMULATION FROM LABEL:

Active Ingredient(s)	% by wt
Thymol (present as a component of Thyme Oil)	0.05%
Other Ingredients	99.95%
Total	100.00%

I BACKGROUND

The product, Benefect Botanical Daily Cleaner Disinfectant Spray (EPA Reg. No. 84683-3), is an EPA-approved disinfectant (bactericide, virucide), sanitizer, and deodorizer for use on hard, non-porous surfaces in household, commercial, institutional, food preparation, animal care, and hospital or medical environments. The applicant requested to amend the registration of this product to add new claims for effectiveness as a fungicide, mildewstat, and carpet sanitizer, as well as claims as a disinfectant against additional microorganisms. The label states that the product is a "one-step" disinfectant. Label directions indicate that the product is effective as a disinfectant and sanitizer on hard, non-porous surfaces in the presence of light to moderate soil loads. Label directions also indicate that the product is effective in sanitizing pre-cleaned carpets. Studies were conducted at ATS Labs, located at 1285 Corporate Center Drive, Suite 110, in Eagan, MN 55121.

This data package contained a letter from the applicant's representative to EPA (dated August 16, 2011), EPA Form 8570-35 (Data Matrix), twenty six studies (MRID 485735-01 through 485735-26), Statements of No Data Confidentiality Claims for all twenty six studies, and the proposed label.

II USE DIRECTIONS

The product is designed for disinfecting and sanitizing hard, non-porous surfaces. The product may be used to treat hard, non-porous surfaces, including: appliance, bathtubs, bed frames, bed rails, bidets, burner trays, cabinets, car interiors, chairs, changing tables, computer screens, computer keyboards, computer mice, coolers, counter tops, cribs, cupboards, cutting boards (plastic), desks, diaper pails, dish racks, door frames, door handles, door knobs, drawers, exhaust fans, faucets, fixtures, floors, foot spas, furniture, garbage cans, grocery carts, gym equipment, hampers, handrails, high chairs, lamps, laundry baskets, light switches, litter boxes, lockers, mirrors, orthotics, outdoor grills, pacifiers, personal protective equipment, pet cages, piano keys, patio furniture, picnic tables and outdoor furniture, playground equipment, prostheses, recycling bins, shelves, shower curtains, shower doors, shower stalls, signs, sinks, sports equipment, storage bins, tables, telephones, television screens, tires, toilet exteriors and seats, tool boxes, toys, vanity tops, wallpaper, walls, waste baskets, water fountains, wheelchairs, window blinds, windows, and work benches. The proposed label indicates that the product may be used on hard, non-porous surfaces, including: baked enamel, Corian, crystal, fiberglass, finished woodwork, glass, glazed ceramic, glazed porcelain, glazed tile, laminated surfaces, linoleum, metal (e.g., aluminum, chrome, stainless steel), painted surfaces, plastic (e.g., vinyl), sealed granite, sealed marble, sealed stone, sealed surfaces, sealed wallboard, and sealed woodwork. Directions on the proposed label provide the following information regarding use of the product:

As a disinfectant: For heavily soiled or greasy areas, a pre-cleaning is required. Wet the surface by sprayer, cloth, sponge, or mop. Leave for 10 minutes. Allow to dry.

As a sanitizer: For heavily soiled or greasy areas, a pre-cleaning is required. Wet the surface by sprayer, cloth, sponge, or mop. Leave for 30 seconds. Allow to air dry.

As a carpet sanitizer: Spray pre-cleaned carpet area until thoroughly wet. Scrub or agitate the carpet as needed to ensure the product is delivered throughout the carpet

pile and backing. Allow the product to remain on the carpet undisturbed for a minimum of 60 minutes.

As a mildewstat: The label does not provide instructions for use of the product against *Aspergillus niger* was demonstrated on pre-cleaned surfaces.

III AGENCY STANDARDS FOR PROPOSED CLAIMS

Disinfectants for Use on Hard Surfaces in Hospital or Medical Environments (Additional Bacteria)

Effectiveness of disinfectants against specific bacteria other than those named in the AOAC Use-Dilution Method, AOAC Germicidal Spray Products as Disinfectants Method, AOAC Fungicidal Test, and AOAC Tuberculocidal Activity Method, must be determined by either the AOAC Use-Dilution Method or the AOAC Germicidal Spray Products as Disinfectants Method. Ten carriers must be tested against each specific microorganism with each of 2 product samples, representing 2 different product lots. To support products labeled as "disinfectants" for specific bacteria (other than those bacteria named in the above test methods), killing of the specific microorganism on all carriers is required.

Disinfectants for Use as Fungicides (Against Pathogenic Fungi, Using the AOAC Germicidal Spray Products as Disinfectants Method)

The effectiveness of liquid disinfectants against specific pathogenic fungi must be supported by efficacy data using an appropriate test. The AOAC Germicidal Spray Products as Disinfectants Method contains procedures for testing fungicidal activity. Ten carriers on each of 2 product samples representing 2 different product lots must be employed in the test. Killing of the specific pathogenic fungi on all carriers is required.

Virucides

The effectiveness of virucides against specific viruses must be supported by efficacy data that simulates, to the extent possible in the laboratory, the conditions under which the product is intended to be used. Carrier methods that are modifications of either the AOAC Use-Dilution Method (for liquid disinfectants) or the AOAC Germicidal Spray Products as Disinfectants Method (for spray disinfectants) must be used. To simulate in-use conditions, the specific virus to be treated must be inoculated onto hard surfaces, allowed to dry, and then treated with the product according to the directions for use on the product label. One surface for each of 2 different product lots of disinfectant must be tested against a recoverable virus titer of at least 10^4 from the test surface for a specified exposure period at room temperature. Then, the virus must be assayed by an appropriate virological technique, using a minimum of four determinations per each dilution assayed. Separate studies are required for each virus. The calculated viral titers must be reported with the test results. For the data to be considered acceptable, results must demonstrate complete inactivation of the virus at all dilutions. When cytotoxicity is evident, at least a 3-log reduction in titer must be demonstrated beyond the cytotoxic level.

Carpet Sanitizers

The effectiveness of sanitizers for carpeting must be supported by data that show that the product will substantially reduce the numbers of test bacteria on the carpeting. Products

that are represented as "one-step sanitizers" should be tested with an appropriate organic soil load. Tests should be performed with each of 3 product samples, representing 3 different product lots, one of which is at least 60 days old against *Staphylococcus aureus* (ATCC 6538) and *Enterobacter aerogenes* (ATCC 13048). The product should be testing using two different types of representative synthetic carpeting, such as acrylic and polypropylene tufted-loop types. If the product is for use in hospital or medical environments, tests should also be performed against *Pseudomonas aeruginosa* (ATCC 15442). If the product is for use on wool carpeting, tests should also be performed on wool carpeting. Results must show a bacterial reduction of at least 99.9 percent over the scrubbed control count.

Mildewstats/Fungistats

The effectiveness of mildewstats and fungistats may be supported by efficacy data derived using the EPA Hard Surface Mildew Fungistatic Test Method. All ten treated tiles must be free of fungal growth after 7 days. To be considered a valid test, untreated control tiles must be at least 50% covered with fungal growth after 7 days. Agency standards are presented in the Pesticide Assessment Guidelines, Subdivision G, Section 93-30, Product Performance, November 1982.

Supplemental Claims

An antimicrobial agent identified as a "one-step" disinfectant or as effective in the presence of organic soil must be tested for efficacy with an appropriate organic soil load, such as 5 percent serum.

IV COMMENTS ON THE SUBMITTED EFFICACY STUDIES

1. MRID 485735-01 "Virucidal Efficacy of a Disinfectant for Use on Inanimate Environmental Surfaces, Virus: Avian Influenza A (H3N2) virus (Avian Reassortant)" for Benefect Botanical Daily Cleaner Disinfectant, by Mary J. Miller. Study conducted at ATS Labs. Study completion date – March 15, 2007. Project Number A04723.

This study was conducted against Avian influenza A (H3N2) virus (Avian Reassortant) (Strain A/Washington/897/80 X A/Mallard/New York/6750/78; ATCC VR-2072), using Rhesus monkey kidney cells (RMK cells; obtained from ViroMed Laboratories, Inc., Cell Culture Division; maintained in-house) as the host system. Two lots (Lot Nos. LC# 071J3012 and LC# 071F1302) of the product, Benefect Botanical Daily Cleaner Disinfectant, were tested according to ATS Labs Protocol No. SLP02012207.AFLU (copy provided). The product was received ready-to-use, as a pump spray. The stock virus culture was adjusted to contain 5% fetal bovine serum as the organic soil load. Films of virus were prepared by spreading 0.2 mL of virus inoculum uniformly over the bottoms of separate sterile glass Petri dishes. The virus films were air-dried for 20 minutes at 19.5°C at 52% relative humidity. For each lot of product, separate dried virus films were sprayed (10 sprays) with the product from a distance of 3-4 inches from the carrier surface until completely covered. The carriers were allowed to remain wet for 10 minutes at 19.5°C. Following exposure, the plates were scraped with a cell scraper to re-suspend the contents. The virus-disinfectant mixtures were passed immediately through individual Sephadex columns, and diluted serially in Minimum Essential Medium with 1% heat-inactivated fetal bovine serum, 10 µg/mL gentamicin, 100 units/mL penicillin, and 2.5 µg/mL amphotericin B. RMK cells in multi-well culture dishes were inoculated in quadruplicate with 0.1 mL of the dilutions. The cultures were incubated at 36-38°C in a humidified atmosphere of 5-

7% CO_2. The cultures were scored periodically for 7 days for the presence or absence of unspecified cytopathic effects, cytotoxicity, and viability. Controls included those for dried virus count, cytotoxicity, and neutralization (both product lots). Viral and cytotoxicity titers were calculated by the method of Spearman Karber.

2. MRID 485735-02 "AOAC Germicidal Spray Method, Test Organism: *Escherichia coli* O157:H7 (ATCC 35150)" for CleanWell Daily Disinfecting Cleaner, by Matthew Sathe. Study conducted at ATS Labs. Study completion date – December 3, 2009. Project Number A08563.

This study was conducted against *Escherichia coli* O157:H7 (ATCC 35150). Two lots (Lot Nos. FM093009003 and FM093009004) of the product, CleanWell Daily Disinfecting Cleaner, were tested using the AOAC Germicidal Spray Products as Disinfectants Method as described in the AOAC Official Methods of Analysis, 17th Edition, 2000. The product was received ready-to-use, as a trigger spray. A culture of the challenge microorganism was prepared in accordance with the published AOAC method. Fetal bovine serum was added to the culture to achieve a 5% organic soil load. Ten glass slide carriers (18 mm x 36 mm) per product lot were inoculated with 10.0 µL of a 48 hour old suspension of test organism. Inoculum was uniformly spread over the entire surface of each carrier. The carriers were dried for 30 minutes at 35-37°C at 40% relative humidity. For each lot of product, separate carriers were sprayed (3 pumps) with the product from a distance of 6-8 inches from the carrier surface until thoroughly wet. The carriers were allowed to remain wet for 10 minutes at 20°C at 23% relative humidity. Following the exposure period, the remaining liquid was drained from each carrier. Individual carriers were transferred to 20 mL of Letheen Broth with 0.07% Lecithin and 0.5% Tween 80 to neutralize. All subcultures were incubated for 48 hours at 35-37°C. Following incubation, the subcultures were examined for the presence or absence of visible growth. Controls included those for carrier population, purity, sterility, viability, and neutralization confirmation (both product lots).

3. MRID 485735-03 "AOAC Germicidal Spray Method, Test Organism: Community Acquired Methicillin Resistant *Staphylococcus aureus* - CA-MRSA (NARSA NRS384)" for CleanWell Daily Disinfectant Cleaner, by Jill Ruhme. Study conducted at ATS Labs. Study completion date – November 23, 2009. Project Number A08445.

This study was conducted against Community Acquired Methicillin Resistant *Staphylococcus aureus* (NARSA NRS384; obtained from the NARSA Contracts Administrator, Focus Technologies, Inc., Herndon, VA). One lot (Lot No. FM093009003) of the product, CleanWell Daily Disinfectant Cleaner, was tested using the AOAC Germicidal Spray Products as Disinfectants Method as described in the AOAC Official Methods of Analysis, 17th Edition, 2000. The product was received ready-to-use, as a trigger spray. A culture of the challenge microorganism was prepared in accordance with the published AOAC method, with the following exception: the culture was incubated for 48-54 hours at 35-37°. Fetal bovine serum was added to the culture to achieve a 5% organic soil load. Ten glass slide carriers (18 mm x 36 mm) were inoculated with 10.0 µL of a 48-54 hour old suspension of test organism. Inoculum was uniformly spread over the entire surface of each carrier. The carriers were dried for 30 minutes at 35-37°C at 40% relative humidity. For the single lot of product, separate carriers were sprayed with the product from a distance of 6 inches from the carrier surface until wet. The carriers were allowed to remain wet for 10 minutes at 20°C at 40% relative humidity. Following the exposure period, the remaining liquid was drained from each carrier. Individual carriers were transferred to 20 mL of Letheen Broth with 0.07% Lecithin and 0.5% Tween 80 to

neutralize. All subcultures were incubated for 48±4 hours at 35-37°C. Following incubation, the subcultures were examined for the presence or absence of visible growth. Controls included those for carrier population, purity, sterility, viability, neutralization confirmation (the single product lot), and antibiotic resistance.

Note The number of sprays was absent from the test method.

Note: Antibiotic resistance of Community Acquired Methicillin Resistant *Staphylococcus aureus* (NARSA NRS384) was verified on a representative culture. The laboratory performed a Kirby Bauer Susceptibility assay. *Staphylococcus aureus* (ATCC 25923) was the control organism. The measured zone of inhibition (i.e., 6 mm) confirmed antibiotic resistance of Community Acquired Methicillin Resistant *Staphylococcus aureus* (NARSA NRS384) to oxacillin. See page 9 and Table 5 of the laboratory report.

4. MRID 485735-04 "AOAC Germicidal Spray Method," Test Organism: Community Acquired Methicillin Resistant *Staphylococcus aureus* - CA-MRSA (NARSA NRS384), for CleanWell Daily Disinfectant Cleaner, by Jill Ruhme. Study conducted at ATS Labs. Study completion date – November 23, 2009. Project Number A08446.

This study was conducted against Community Acquired Methicillin Resistant *Staphylococcus aureus* (NARSA NRS384; obtained from the NARSA Contracts Administrator, Focus Technologies, Inc., Herndon, VA). One lot (Lot No. FM093009004) of the product, CleanWell Daily Disinfectant Cleaner, was tested using the AOAC Germicidal Spray Products as Disinfectants Method as described in the AOAC Official Methods of Analysis, 17th Edition, 2000. The product was received ready-to-use, as a trigger spray. A culture of the challenge microorganism was prepared in accordance with the published AOAC method, with the following exception: the culture was incubated for 48-54 hours at 35-37°C. Fetal bovine serum was added to the culture to achieve a 5% organic soil load. Ten glass slide carriers (18 mm x 36 mm) were inoculated with 10.0 µL of a 48-54 hour old suspension of test organism. Inoculum was uniformly spread over the entire surface of each carrier. The carriers were dried for 30 minutes at 35-37°C at 40% relative humidity. For the single lot of product, separate carriers were sprayed with the product from a distance of 6 inches from the carrier surface until wet. The carriers were allowed to remain wet for 10 minutes at 20°C at 25% relative humidity. Following the exposure period, the remaining liquid was drained from each carrier. Individual carriers were transferred to 20 mL of Letheen Broth with 0.07% Lecithin and 0.5% Tween 80 to neutralize. All subcultures were incubated for 48±4 hours at 35-37°C. Following incubation, the subcultures were examined for the presence or absence of visible growth. Controls included those for carrier population, purity, sterility, viability, neutralization confirmation (the single product lot), and antibiotic resistance.

Note The number of sprays was absent from the test method.

Note: Antibiotic resistance of Community Acquired Methicillin Resistant *Staphylococcus aureus* (NARSA NRS384) was verified on a representative culture. The laboratory performed a Kirby Bauer Susceptibility assay. *Staphylococcus aureus* (ATCC 25923) was the control organism. The measured zone of inhibition (i.e., 6 mm) confirmed antibiotic resistance of Community Acquired Methicillin Resistant *Staphylococcus aureus* (NARSA NRS384) to oxacillin. See page 9 and Table 5 of the laboratory report.

5. MRID 485735-05 "Fungicidal Germicidal Spray Method, Test Organism: *Trichophyton mentagrophytes* (ATCC 9533)" for Benefect Daily Disinfectant Cleaner, by Matthew Sathe. Study conducted at ATS Labs. Study completion date – March 2, 2010. Project Number A08979.

This study was conducted against *Trichophyton mentagrophytes* (ATCC 9533). One lot (Lot No. D089662) of the product, Benefect Daily Disinfectant Cleaner, was tested using the AOAC Germicidal Spray Products as Disinfectants Method as described in the AOAC Official Methods of Analysis, 17th Edition, 2000. The product was received ready-to-use, as a trigger spray. A culture of the challenge microorganism was prepared. Fetal bovine serum was added to the conidial suspension to achieve a 5% organic soil load. Ten glass slide carriers (18 mm x 36 mm) were inoculated with 0.01 mL of a 10 day old suspension of test organism. The inoculum was uniformly spread over the surface of each carrier. The carriers were dried for 30 minutes at 35-37°C at 39% humidity. For the single lot of product, separate carriers were sprayed with the product from a distance of 6-8 inches from the carrier surface until thoroughly wet. The carriers were allowed to remain wet for 10 minutes at 22.8°C at 10.0% relative humidity. Following the exposure period, the remaining liquid was drained from each carrier. Individual carriers were transferred to 20 mL of Sabouraud Dextrose Broth with 0.07% Lecithin and 0.5% Tween 80 to neutralize. All subcultures were incubated for 10 days at 25-30°C. The subcultures were stored for 1 day at 2-8°C prior to examination. Following incubation and storage, the subcultures were examined for the presence or absence of visible growth. Controls included those for carrier population, purity, sterility, viability, and neutralization confirmation (the single product lot).

6. MRID 485735-06 "Fungicidal Germicidal Spray Method, Test Organism: *Trichophyton mentagrophytes* (ATCC 9533)" for Benefect Daily Disinfectant Cleaner, by Matthew Sathe. Study conducted at ATS Labs. Study completion date – March 2, 2010. Project Number A08980.

This study was conducted against *Trichophyton mentagrophytes* (ATCC 9533). One lot (Lot No. D119725) of the product, Benefect Daily Disinfectant Cleaner, was tested using the AOAC Germicidal Spray Products as Disinfectants Method as described in the AOAC Official Methods of Analysis, 17th Edition, 2000. The product was received ready-to-use, as a trigger spray. A culture of the challenge microorganism was prepared. Fetal bovine serum was added to the conidial suspension to achieve a 5% organic soil load. Ten glass slide carriers (18 mm x 36 mm) were inoculated with 0.01 mL of a 10 day old suspension of test organism. The inoculum was uniformly spread over the surface of each carrier. The carriers were dried for 30 minutes at 35-37°C at 39% humidity. For the single lot of product, separate carriers were sprayed with the product from a distance of 6-8 inches from the carrier surface until thoroughly wet. The carriers were allowed to remain wet for 10 minutes at 22.8°C at 10.0% relative humidity. Following the exposure period, the remaining liquid was drained from each carrier. Individual carriers were transferred to 20 mL of Sabouraud Dextrose Broth with 0.07% Lecithin and 0.5% Tween 80 to neutralize. All subcultures were incubated for 10 days at 25-30°C. The subcultures were stored for 1 day at 2-8°C prior to examination. Following incubation and storage, the subcultures were examined for the presence or absence of visible growth. Controls included those for carrier population, purity, sterility, viability, and neutralization confirmation (the single product lot).

7. MRID 485735-07 "EPA Hard Surface Mildew-Fungistatic Test, Test Organism: *Aspergillus niger* (ATCC 6275)" for Benefect Daily Disinfectant Cleaner, by Becky Lien. Study conducted at ATS Labs. Study completion date – November 22, 2010. Project Number A10417.

This study was conducted against *Aspergillus niger* (ATCC 6275). One lot (Lot No. FM100710001) of the product, Benefect Daily Disinfectant Cleaner, was tested using the EPA Hard Surface Mildew Fungistatic Test Method. The product was received ready-to-use. A culture of the challenge microorganism was prepared in accordance with the EPA method, with the following exceptions: (1) the culture was incubated for 10 days at 25-30°C; and (2) sterile glass beads and saline/Triton solution were added to the flask, and the flask was agitated A 1.00 mL aliquot of a standardized conidial suspension was added to 20.0 mL of sterile Czapek's solution. The product was not tested in the presence of a 5% organic soil load. Ten sterile 1 inch x 1 inch glazed ceramic tiles were used. The tiles were sprayed (12 sprays) with the product at a distance of 4-6 inches until thoroughly wet, immediately removed, and placed in a vertical or near vertical position to allow excess liquid to drain. Following treatment, the tiles were dried for 24 minutes at 35-37°C. Following the drying period, the surfaces of each test tile and each untreated control tile were sprayed with the *Aspergillus niger* conidia-Czapek suspension using a DeVilbiss #151 Atomizer. The tiles were returned to a 35-37°C incubator and dried for 41 minutes. Each tile (treated side up) was placed in an individual Petri dish containing hardened sterile water agar. The plates were incubated for 7 days at 25-30°C at a minimum of 95% relative humidity. The tiles were examined for the presence or absence of fungal growth after 7 days of incubation. When no growth was visually observed, a magnified examination was performed. Controls included those for purity and sterility.

8. MRID 485735-08 "EPA Hard Surface Mildew-Fungistatic Test, Test Organism: *Aspergillus niger* (ATCC 6275)" for Benefect Daily Disinfectant Cleaner, by Becky Lien. Study conducted at ATS Labs. Study completion date – November 22, 2010. Project Number A10418.

This study was conducted against *Aspergillus niger* (ATCC 6275). One lot (Lot No. FM100710002) of the product, Benefect Daily Disinfectant Cleaner, was tested using the EPA Hard Surface Mildew Fungistatic Test Method. The product was received ready-to-use. A culture of the challenge microorganism was prepared. A 1.00 mL aliquot of a standardized conidial suspension was added to 20.0 mL of sterile Czapek's solution. The product was not tested in the presence of a 5% organic soil load. Ten sterile 1 inch x 1 inch glazed ceramic tiles were used. The tiles were sprayed (12 sprays) with the product at a distance of 4-6 inches until wet, immediately removed, and placed in a vertical or near vertical position to allow excess liquid to drain. Following treatment, the tiles were dried for 24 minutes at 35-37°C. Following the drying period, the surfaces of each test tile and each untreated control tile were sprayed with the *Aspergillus niger* conidia-Czapek suspension using a DeVilbiss #151 Atomizer. The tiles were returned to a 35-37°C incubator and dried for 38 minutes. Each tile (treated side up) was placed in an individual Petri dish containing hardened sterile water agar. The plates were incubated for 7 days at 25-30°C at a minimum of 95% relative humidity. The tiles were examined for the presence or absence of fungal growth after 7 days of incubation. When no growth was visually observed, a magnified examination was performed. Controls included those for purity and sterility.

9. MRID 485735-09 "Carpet Sanitizer," Test Organism: *Staphylococcus aureus* (ATCC 6538), for Benefect Daily Disinfectant-Cleaner, by Matthew Sathe. Study conducted at ATS Labs. Study completion date – July 9, 2010. Project Number A09634.

This study was conducted against *Staphylococcus aureus* (ATCC 6538). One lot (Lot No. FM040210001) of the product, Benefect Daily Disinfectant-Cleaner, was tested. The laboratory report referenced the Carpet Sanitizer Test from DIS/TSS-8. The product lot tested was at least 60 days old at the time of testing. The product was received ready-to-use. A culture of the challenge microorganism was prepared in accordance with the EPA method. The inoculum suspension was adjusted to a 4.0 McFarland turbidity standard to produce an average of ~1.0 x 10^9 CFU/mL. The product was not tested in the presence of a 5% organic soil load. Nylon carpet was cut into 8 inch x 12 inch carpet pieces, and six 2 inch x 2 inch square carriers were cut into the larger carpet pieces. The carpet was fastened to a mounting tray and sterilized by autoclave. Each cut square carrier was inoculated with 100 µL of an 18-24 hour old suspension of test organism. The inoculum was spread evenly over the surface of each cut square carrier. The cut square carriers were dried in an incubator for 60 minutes at 35-37°C with sterile foil loosely covering the top. For the single lot of product, separate cut square carriers were sprayed (15 pumps) with the product from a distance of ~6 inches from the carrier surface. Each cut square carrier was then scrubbed for 30 seconds using 30 circular clockwise strokes and 30 circular counterclockwise strokes. A circular area of pile ~3 inches in diameter around the center of each cut square carrier was scrubbed. A new brush was used for each cut square carrier. The cut square carriers remained at 23.3°C for 60 minutes. Following exposure, each cut square carrier was removed from the larger carpet piece and transferred to a sterile vessel containing 100 mL of Letheen Broth with 0.07% Lecithin and 0.5% Tween 80 and 10 stainless steel penicylinders. Each vessel was shaken vigorously for 1 minute at 200 RPM to free the bacteria from the carpet fibers. Ten-fold serial dilutions were prepared and 1.00 mL of the 10^0 through 10^{-3} dilutions were plated in duplicate on tryptic soy agar with 5% sheep's blood. All subcultures were incubated for 44 hours at 35-37°C. The subcultures were stored for 2 days at 2-8°C prior to examination. Following incubation and storage, the subcultures were enumerated. Controls included those for unscrubbed population count, scrubbed population count, purity, sterility, viability, and neutralization confirmation (the single product lot).

10. MRID 485735-10 "Carpet Sanitizer, Test Organism: *Staphylococcus aureus* (ATCC 6538)" for Benefect Daily Disinfectant-Cleaner, by Becky Lien. Study conducted at ATS Labs. Study completion date – July 7, 2010. Amended report date – September 9, 2010. Project Number A09635.

This study was conducted against *Staphylococcus aureus* (ATCC 6538). One lot (Lot No. D089662) of the product, Benefect Daily Disinfectant-Cleaner, was tested. The laboratory report referenced the Carpet Sanitizer Test from DIS/TSS-8. The product lot tested was at least 60 days old at the time of testing. The product was received ready-to-use. A culture of the challenge microorganism was prepared in accordance with the EPA method (which references AOAC Method 960.09D). The inoculum suspension was adjusted to a 4.0 McFarland turbidity standard to produce an average of ~1.0 x 10^9 CFU/mL. The product was not tested in the presence of a 5% organic soil load. Nylon carpet was cut into 8 inch x 12 inch carpet pieces, and six 2 inch x 2 inch square carriers were cut into the larger carpet pieces. The carpet was fastened to a mounting tray and sterilized by autoclave. Each cut square carrier was inoculated

with 100 µL of an 18-24 hour old suspension of test organism. The inoculum was spread evenly over the surface of each cut square carrier. The cut square carriers were dried in an incubator for 60 minutes at 35-37°C. For the single lot of product, separate cut square carriers were sprayed (presumably 15-20 pumps) with the product (presumably from a distance of 6 inches from the carrier surface). Each cut square carrier was then scrubbed for 30 seconds using 30 circular clockwise strokes and 30 circular counterclockwise strokes. A circular area of pile ~3 inches in diameter around the center of each cut square carrier was scrubbed. A new brush was used for each cut square carrier. The cut square carriers remained at 23.0°C for 60 minutes. Following exposure, each cut square carrier was removed from the larger carpet piece and transferred to a sterile vessel containing 100 mL of Letheen Broth with 0.07% Lecithin and 0.5% Tween 80 and 10 stainless steel penicylinders. Each vessel was shaken vigorously for 1 minute at 200 RPM to free the bacteria from the carpet fibers. Ten-fold serial dilutions were prepared and 1.00 mL of the 10^0 through 10^{-3} dilutions were plated in duplicate on tryptic soy agar with 5% sheep's blood. All subcultures were incubated for ~44 hours at 35-37°C. The subcultures were stored for 2 days at 2-8°C prior to examination. Following incubation and storage, the subcultures were enumerated. Controls included those for unscrubbed population count, scrubbed population count, purity, sterility, viability, and neutralization confirmation (the single product lot).

11. MRID 485735-11 "Carpet Sanitizer, Test Organism: *Staphylococcus aureus* (ATCC 6538)" for Benefect Daily Disinfectant-Cleaner, by Matthew Sathe. Study conducted at ATS Labs. Study completion date – July 9, 2010. Project Number A09636.

This study was conducted against *Staphylococcus aureus* (ATCC 6538). One lot (Lot No. D119725) of the product, Benefect Daily Disinfectant-Cleaner, was tested. The laboratory report referenced the Carpet Sanitizer Test from DIS/TSS-8. The product lot tested was at least 60 days old at the time of testing. The product was received ready-to-use. A culture of the challenge microorganism was prepared in accordance with the EPA method (which references AOAC Method 960.09D). The inoculum suspension was adjusted to a 4.0 McFarland turbidity standard to produce an average of ~1.0×10^9 CFU/mL. The product was not tested in the presence of a 5% organic soil load. Nylon carpet was cut into 8 inch x 12 inch carpet pieces, and six 2 inch x 2 inch square carriers were cut into the larger carpet pieces. The carpet was fastened to a mounting tray and sterilized by autoclave. Each cut square carrier was inoculated with 100 µL of an 18-24 hour old suspension of test organism. The inoculum was spread evenly over the surface of each cut square carrier. The cut square carriers were dried in an incubator for 60 minutes at 35-37°C with sterile foil loosely covering the top. For the single lot of product, separate cut square carriers were sprayed (15 pumps) with the product from a distance of ~6 inches from the carrier surface. Each cut square carrier was then scrubbed for 30 seconds using 30 circular clockwise strokes and 30 circular counterclockwise strokes. A circular area of pile ~3 inches in diameter around the center of each cut square carrier was scrubbed. A new brush was used for each cut square carrier. The cut square carriers remained at 22.3°C for 60 minutes. Following exposure, each cut square carrier was removed from the larger carpet piece and transferred to a sterile vessel containing 100 mL of Letheen Broth with 0.07% Lecithin and 0.5% Tween 80 and 10 stainless steel penicylinders. Each vessel was shaken vigorously for 1 minute at 200 RPM to free the bacteria from the carpet fibers. Ten-fold serial dilutions were prepared and 1.00 mL of the 10^0 through 10^{-3} dilutions were plated in duplicate on tryptic soy agar with 5% sheep's blood. All subcultures were incubated for ~44 hours at 35-37°C. The subcultures were stored for 2 days at 2-8°C prior to examination. Following incubation and storage, the subcultures were enumerated. Controls included those for unscrubbed population

count, scrubbed population count, purity, sterility, viability, and neutralization confirmation (the single product lot).

12. MRID 485735-12 "Carpet Sanitizer, Test Organism: *Staphylococcus aureus* (ATCC 6538)" for Benefect Daily Disinfectant-Cleaner, by Becky Lien. Study conducted at ATS Labs. Study completion date – December 27, 2010. Project Number A10625.

This study was conducted against *Staphylococcus aureus* (ATCC 6538). One lot (Lot No. FM100710001) of the product, Benefect Daily Disinfectant-Cleaner, was tested. The laboratory report referenced the Carpet Sanitizer Test from DIS/TSS-8. The product lot tested was at least 60 days old at the time of testing. The product was received ready-to-use, as a trigger spray. A culture of the challenge microorganism was prepared in accordance with the EPA method (which references AOAC Method 960.09D). The inoculum suspension density was comparable to 1 x 10^9 CFU/mL; no further adjustment was necessary. The product was not tested in the presence of a 5% organic soil load. Polypropylene/Olefin carpet was cut into 8 inch x 12 inch carpet pieces, and six 2 inch x 2 inch square carriers were cut into the larger carpet pieces. The carpet was fastened to a mounting tray and sterilized by autoclave. Each cut square carrier was inoculated with 100 µL of an 18-24 hour old suspension of test organism. The inoculum was spread evenly over the surface of each cut square carrier. The cut square carriers were dried in an incubator for 60 minutes at 35-37°C with sterile foil loosely covering the top. For the single lot of product, separate cut square carriers were sprayed (16 pumps) with the product from a distance of ~4-6 inches from the carrier surface. Each cut square carrier was then scrubbed for 30 seconds using 30 circular clockwise strokes and 30 circular counterclockwise strokes. A circular area of pile ~3 inches in diameter around the center of each cut square carrier was scrubbed. A new brush was used for each cut square carrier. The cut square carriers remained at 23.0°C for 60 minutes. Following exposure, each cut square carrier was removed from the larger carpet piece and transferred to a sterile vessel containing 100 mL of Letheen Broth with 0.07% Lecithin and 0.5% Tween 80 and 10 stainless steel penicylinders. Each vessel was shaken vigorously for 1 minute at ~200 RPM to free the bacteria from the carpet fibers. Ten-fold serial dilutions were prepared and 1.00 mL of the 10^0 through 10^{-3} dilutions were plated in duplicate on tryptic soy agar with 5% sheep's blood. All subcultures were incubated for 47.5 hours at 35-37°C. The subcultures were stored for 1 day at 2-8°C prior to examination. Following incubation and storage, the subcultures were enumerated. Controls included those for unscrubbed population count, scrubbed population count, purity, sterility, and neutralization confirmation (the single product lot).

13. MRID 485735-13 "Carpet Sanitizer, Test Organism: *Staphylococcus aureus* (ATCC 6538)" for Benefect Daily Disinfectant-Cleaner, by Becky Lien. Study conducted at ATS Labs. Study completion date – January 10, 2011. Project Number A10697.

This study was conducted against *Staphylococcus aureus* (ATCC 6538). One lot (Lot No. FM100710002) of the product, Benefect Daily Disinfectant-Cleaner, was tested. The laboratory report referenced the Carpet Sanitizer Test from DIS/TSS-8. The product lot tested was at least 60 days old at the time of testing. The product was received ready-to-use, as a trigger spray. A culture of the challenge microorganism was prepared in accordance with the EPA method (which references AOAC Method 960.09D). The inoculum suspension had a final spectrophotometer value of 2.197. The product was not tested in the presence of a 5% organic

soil load. Olefin carpet was cut into 8 inch x 12 inch carpet pieces, and six 2 inch x 2 inch square carriers were cut into the larger carpet pieces. The carpet was fastened to a mounting tray and sterilized by autoclave. Each cut square carrier was inoculated with 100 µL of an 18-24 hour old suspension of test organism. The inoculum was spread evenly over the surface of each cut square carrier. The cut square carriers were dried in an incubator for 60 minutes at 35-37°C with sterile foil loosely covering the top. For the single lot of product, separate cut square carriers were sprayed (16 pumps) with the product from a distance of 4-6 inches from the carrier surface. Each cut square carrier was then scrubbed for 30 seconds using 30 circular clockwise strokes and 30 circular counterclockwise strokes. A circular area of pile ~3 inches in diameter around the center of each cut square carrier was scrubbed. A new brush was used for each cut square carrier. The cut square carriers remained at 23.5°C for 60 minutes. Following exposure, each cut square carrier was removed from the larger carpet piece and transferred to a sterile vessel containing 100 mL of Letheen Broth with 0.07% Lecithin and 0.5% Tween 80 and 10 stainless steel penicylinders. Each vessel was shaken vigorously for 1 minute at 200 RPM to free the bacteria from the carpet fibers. Ten-fold serial dilutions were prepared and 1.00 mL of the 10^0 through 10^{-3} dilutions were plated in duplicate on tryptic soy agar with 5% sheep's blood. All subcultures were incubated for 45 hours at 35-37°C. The subcultures were stored for 2 days at 2-8°C prior to examination. Following incubation and storage, the subcultures were enumerated. Controls included those for unscrubbed population count, scrubbed population count, purity, sterility, and neutralization confirmation (the single product lot).

14. MRID 485735-14 "Carpet Sanitizer, Test Organism: *Staphylococcus aureus* (ATCC 6538)" for Benefect Daily Disinfectant-Cleaner, by Becky Lien. Study conducted at ATS Labs. Study completion date – January 10, 2011. Project Number A10698.

This study was conducted against *Staphylococcus aureus* (ATCC 6538). One lot (Lot No. FM040210001) of the product, Benefect Daily Disinfectant-Cleaner, was tested. The laboratory report referenced the Carpet Sanitizer Test from DIS/TSS-8. The product lot tested was at least 60 days old at the time of testing. The product was received ready-to-use, as a trigger spray. A culture of the challenge microorganism was prepared in accordance with the EPA method (which references AOAC Method 960.09D). The inoculum suspension had a final spectrophotometer value of 2.197. The product was not tested in the presence of a 5% organic soil load. Olefin carpet was cut into 8 inch x 12 inch carpet pieces, and six 2 inch x 2 inch square carriers were cut into the larger carpet pieces. The carpet was fastened to a mounting tray and sterilized by autoclave. Each cut square carrier was inoculated with 100 µL of an 18-24 hour old suspension of test organism. The inoculum was spread evenly over the surface of each cut square carrier. The cut square carriers were dried in an incubator for 60 minutes at 35-37°C with sterile foil loosely covering the top. For the single lot of product, separate cut square carriers were sprayed (16 pumps) with the product from a distance of 4-6 inches from the carrier surface. Each cut square carrier was then scrubbed for 30 seconds using 30 circular clockwise strokes and 30 circular counterclockwise strokes. A circular area of pile ~3 inches in diameter around the center of each cut square carrier was scrubbed. A new brush was used for each cut square carrier. The cut square carriers remained at 23.0°C for 60 minutes. Following exposure, each cut square carrier was removed from the larger carpet piece and transferred to a sterile vessel containing 100 mL of Letheen Broth with 0.07% Lecithin and 0.5% Tween 80 and 10 stainless steel penicylinders. Each vessel was shaken vigorously for 1 minute at 200 RPM to free the bacteria from the carpet fibers. Ten-fold serial dilutions were prepared and 1.00 mL of the 10^0 through 10^{-3} dilutions were plated in duplicate on tryptic soy agar with 5% sheep's blood. All subcultures were incubated for 44.75 hours at 35-37°C. The subcultures

were stored for 3 days at 2-8°C prior to examination. Following incubation and storage, the subcultures were enumerated. Controls included those for unscrubbed population count, scrubbed population count, purity, sterility, and neutralization confirmation (the single product lot).

15. MRID 485735-15 "Carpet Sanitizer, Test Organism: *Enterobacter aerogenes* (ATCC 13048)" for Benefect Daily Disinfectant-Cleaner, by Jill Ruhme. Study conducted at ATS Labs. Study completion date – September 2, 2010. Project Number A09872.

This study was conducted against *Enterobacter aerogenes* (ATCC 13048). One lot (Lot No. FM040210001) of the product, Benefect Daily Disinfectant-Cleaner, was tested. The laboratory report referenced the Carpet Sanitizer Test from DIS/TSS-8. The product lot tested was at least 60 days old at the time of testing (based on information provided in MRID 485735-09). The product was received ready-to-use, as a trigger spray. A culture of the challenge microorganism was prepared in accordance with the EPA method (which references AOAC Method 960.09D). The inoculum suspension was compared to a 4.0 McFarland turbidity standard to produce an average of ~1.0 x 10^9 CFU/mL. The product was not tested in the presence of a 5% organic soil load. Nylon carpet was cut into 8 inch x 12 inch carpet pieces, and six 2 inch x 2 inch square carriers were cut into the larger carpet pieces. The carpet was fastened to a mounting tray and sterilized by autoclave. Each cut square carrier was inoculated with 100 µL of an 18-24 hour old suspension of test organism. The inoculum was spread evenly over the surface of each cut square carrier. The cut square carriers were dried in an incubator for 60 minutes at 35-37°C with sterile foil loosely covering the top. For the single lot of product, separate cut square carriers were sprayed (15 pumps) with the product from a distance of ~6 inches from the carrier surface until thoroughly wet. Each cut square carrier was then scrubbed for 30 seconds using 30 circular clockwise strokes and 30 circular counterclockwise strokes. A circular area of pile ~3 inches in diameter around the center of each cut square carrier was scrubbed. A new brush was used for each cut square carrier. The cut square carriers remained at 21.5°C for 60 minutes. Following exposure, each cut square carrier was removed from the larger carpet piece and transferred to a sterile vessel containing 100 mL of Letheen Broth with 0.07% Lecithin and 0.5% Tween 80 and 10 stainless steel penicylinders. Each vessel was shaken vigorously for 1 minute at 200 RPM to free the bacteria from the carpet fibers. Ten-fold serial dilutions were prepared and 1.00 mL of the 10^0 through 10^{-3} dilutions were plated in duplicate on tryptic soy agar with 5% sheep's blood. All subcultures were incubated for 45.75 hours at 25-30°C. Following incubation, the subcultures were enumerated. Controls included those for unscrubbed population count, scrubbed population count, purity, sterility, viability, and neutralization confirmation (the single product lot).

16. MRID 485735-16 "Carpet Sanitizer, Test Organism: *Enterobacter aerogenes* (ATCC 13048)" for Benefect Daily Disinfectant-Cleaner, by Becky Lien. Study conducted at ATS Labs. Study completion date – December 27, 2010. Project Number A10630.

This study was conducted against *Enterobacter aerogenes* (ATCC 13048). One lot (Lot No. FM100710001) of the product, Benefect Daily Disinfectant-Cleaner, was tested. The laboratory report referenced the Carpet Sanitizer Test from DIS/TSS-8. The product lot tested was at least 60 days old at the time of testing. The product was received ready-to-use, as a

trigger spray. A culture of the challenge microorganism was prepared in accordance with the EPA method (which references AOAC Method 960.09D). The inoculum suspension density was comparable to ~1.0 x 10^9 CFU/mL; no further adjustment was necessary. The product was not tested in the presence of a 5% organic soil load. Nylon carpet was cut into 8 inch x 12 inch carpet pieces, and six 2 inch x 2 inch square carriers were cut into the larger carpet pieces. The carpet was fastened to a mounting tray and sterilized by autoclave. Each cut square carrier was inoculated with 100 µL of a ~23 hour old suspension of test organism. The inoculum was spread evenly over the surface of each cut square carrier. The cut square carriers were dried in an incubator for 60 minutes at 35-37°C with sterile foil loosely covering the top. For the single lot of product, separate cut square carriers were sprayed (16 pumps) with the product from a distance of ~4-6 inches from the carrier surface. Each cut square carrier was then scrubbed for 30 seconds using 30 circular clockwise strokes and 30 circular counterclockwise strokes. A circular area of pile ~3 inches in diameter around the center of each cut square carrier was scrubbed. A new brush was used for each cut square carrier. The cut square carriers remained at 20.9°C for 60 minutes. Following exposure, each cut square carrier was removed from the larger carpet piece and transferred to a sterile vessel containing 100 mL of Letheen Broth with 0.07% Lecithin and 0.5% Tween 80 and 10 stainless steel penicylinders. Each vessel was shaken vigorously for 1 minute at ~200 RPM to free the bacteria from the carpet fibers. Ten-fold serial dilutions were prepared and 1.00 mL of the 10^0 through 10^{-3} dilutions were plated in duplicate on tryptic soy agar with 5% sheep's blood. All subcultures were incubated for 44.5 hours at 35-37°C. Following incubation, the subcultures were enumerated. Controls included those for unscrubbed population count, scrubbed population count, purity, sterility, and neutralization confirmation (the single product lot).

17. MRID 485735-17 "Carpet Sanitizer, Test Organism: *Enterobacter aerogenes* (ATCC 13048)" for Benefect Daily Disinfectant-Cleaner, by Becky Lien. Study conducted at ATS Labs. Study completion date – February 7, 2011. Project Number A10837.

This study was conducted against *Enterobacter aerogenes* (ATCC 13048). One lot (Lot No. Test010511-001 (i.e., Lot FM100710002)) of the product, Benefect Daily Disinfectant-Cleaner, was tested. The laboratory report referenced the Carpet Sanitizer Test from DIS/TSS-8. The product was received ready-to-use, as a trigger spray. A culture of the challenge microorganism was prepared in accordance with the EPA method (which references AOAC Method 960.09D). The inoculum had a final spectrophotometer value (at 620 nm) of 0.895. The product was not tested in the presence of a 5% organic soil load. Nylon carpet was cut into 8 inch x 12 inch carpet pieces, and six 2 inch x 2 inch square carriers were cut into the larger carpet pieces. The carpet was fastened to a mounting tray and sterilized by autoclave. Each cut square carrier was inoculated with 100 µL of an 18-24 hour old suspension of test organism. The inoculum was spread evenly over the surface of each cut square carrier. The cut square carriers were dried in an incubator for 60 minutes at 35-37°C with sterile foil loosely covering the top. For the single lot of product, separate cut square carriers were sprayed (20 pumps) with the product from a distance of 4-6 inches from the carrier surface. Each cut square carrier was then scrubbed for 30 seconds using 30 circular clockwise strokes and 30 circular counterclockwise strokes. A circular area of pile ~3 inches in diameter around the center of each cut square carrier was scrubbed. A new brush was used for each cut square carrier. The cut square carriers remained at 23.1°C for 60 minutes. Following exposure, each cut square carrier was removed from the larger carpet piece and transferred to a sterile vessel containing 100 mL of Letheen Broth with 0.07% Lecithin and 0.5% Tween 80 and 10 stainless steel penicylinders. Each vessel was shaken for 1 minute at 200 RPM to free the bacteria from the

carpet fibers. Ten-fold serial dilutions were prepared and 1.00 mL of the 10^0 through 10^{-3} dilutions were plated in duplicate on tryptic soy agar with 5% sheep's blood. All subcultures were incubated for 44 hours at 25-30°C. Following incubation, the subcultures were enumerated. Controls included those for unscrubbed population count, scrubbed population count, purity, sterility, and neutralization confirmation (the single product lot).

18. MRID 485735-18 "Carpet Sanitizer, Test Organism: *Enterobacter aerogenes* (ATCC 13048)" for Benefect Daily Disinfectant-Cleaner, by Lynsey Wieland. Study conducted at ATS Labs. Study completion date – December 9, 2010. Project Number A10542.

This study was conducted against *Enterobacter aerogenes* (ATCC 13048). One lot (Lot No. FM100710001) of the product, Benefect Daily Disinfectant-Cleaner, was tested. The laboratory report referenced the Carpet Sanitizer Test from DIS/TSS-8. The product was received ready-to-use, as a trigger spray. A culture of the challenge microorganism was prepared in accordance with the EPA method (which references AOAC Method 960.09D). The inoculum suspension appeared appropriately turbid to produce an average of ~1.0×10^9 CFU/mL. The product was not tested in the presence of a 5% organic soil load. Polypropylene/Olefin carpet was cut into 8 inch x 12 inch carpet pieces, and six 2 inch x 2 inch square carriers were cut into the larger carpet pieces. The carpet was fastened to a mounting tray and sterilized by autoclave. Each cut square carrier was inoculated with 100 µL of a 24 hour old suspension of test organism. The inoculum was spread evenly over the surface of each cut square carrier. The cut square carriers were dried in an incubator for 60 minutes at 35-37°C with sterile foil loosely covering the top. For the single lot of product, separate cut square carriers were sprayed (16 pumps) with the product from a distance of ~4-6 inches from the carrier surface. Each cut square carrier was then scrubbed for 30 seconds using 30 circular clockwise strokes and 30 circular counterclockwise strokes. A circular area of pile ~3 inches in diameter around the center of each cut square carrier was scrubbed. A new brush was used for each cut square carrier. The cut square carriers remained at 24.4°C for 60 minutes. Following exposure, each cut square carrier was removed from the larger carpet piece and transferred to a sterile vessel containing 100 mL of Letheen Broth with 0.07% Lecithin and 0.5% Tween 80 and 10 stainless steel penicylinders. Each vessel was shaken vigorously for 1 minute at 200 RPM to free the bacteria from the carpet fibers. Ten-fold serial dilutions were prepared and 1.00 mL of the 10^0 through 10^{-3} dilutions were plated in duplicate on tryptic soy agar with 5% sheep's blood. All subcultures were incubated for 44 hours at 25-30°C. The subcultures were stored for 2 days at 2-8°C prior to examination. Following incubation and storage, the subcultures were enumerated. Controls included those for unscrubbed population count, scrubbed population count, purity, sterility, and neutralization confirmation (the single product lot).

19. MRID 485735-19 "Carpet Sanitizer, Test Organism: *Enterobacter aerogenes* (ATCC 13048)" for Benefect Daily Disinfectant-Cleaner, by Lynsey Wieland. Study conducted at ATS Labs. Study completion date – December 29, 2010. Project Number A10628.

This study was conducted against *Enterobacter aerogenes* (ATCC 13048). One lot (Lot No. FM100710002) of the product, Benefect Daily Disinfectant-Cleaner, was tested. The laboratory report referenced the Carpet Sanitizer Test from DIS/TSS-8. The product lot tested was at least 60 days old at the time of testing. The product was received ready-to-use, as a

trigger spray. A culture of the challenge microorganism was prepared in accordance with the EPA method (which references AOAC Method 960.09D). The inoculum suspension appeared appropriately turbid to produce an average of ~1 x 10^9 CFU/mL. The product was not tested in the presence of a 5% organic soil load. Polypropylene/Olefin carpet was cut into 8 inch x 12 inch carpet pieces, and six 2 inch x 2 inch square carriers were cut into the larger carpet pieces. The carpet was fastened to a mounting tray and sterilized by autoclave. Each cut square carrier was inoculated with 100 µL of a 22.75 hour old suspension of test organism. The inoculum was spread evenly over the surface of each cut square carrier. The cut square carriers were dried in an incubator for 60 minutes at 35-37°C with sterile foil loosely covering the top. For the single lot of product, separate cut square carriers were sprayed (16 pumps) with the product from a distance of 4-6 inches from the carrier surface. Each cut square carrier was then scrubbed for 30 seconds using 30 circular clockwise strokes and 30 circular counterclockwise strokes. A circular area of pile ~3 inches in diameter around the center of each cut square carrier was scrubbed. A new brush was used for each cut square carrier. The cut square carriers remained at 22.8°C for 60 minutes. Following exposure, each cut square carrier was removed from the larger carpet piece and transferred to a sterile vessel containing 100 mL of Letheen Broth with 0.07% Lecithin and 0.5% Tween 80 and 10 stainless steel penicylinders. Each vessel was shaken vigorously for 1 minute at 200 RPM to free the bacteria from the carpet fibers. Ten-fold serial dilutions were prepared and 1.00 mL of the 10^0 through 10^{-3} dilutions were plated in duplicate on tryptic soy agar with 5% sheep's blood. All subcultures were incubated for 45 hours at 25-30°C. Following incubation, the subcultures were enumerated. Controls included those for unscrubbed population count, scrubbed population count, purity, sterility, and neutralization confirmation (the single product lot).

20. MRID 485735-20 "Carpet Sanitizer, Test Organism: *Enterobacter aerogenes* (ATCC 13048)" for Benefect Daily Disinfectant-Cleaner, by Lynsey Wieland. Study conducted at ATS Labs. Study completion date – December 29, 2010. Project Number A10629.

This study was conducted against *Enterobacter aerogenes* (ATCC 13048). One lot (Lot No. FM040210001) of the product, Benefect Daily Disinfectant-Cleaner, was tested. The laboratory report referenced the Carpet Sanitizer Test from DIS/TSS-8. The product lot tested was at least 60 days old at the time of testing. The product was received ready-to-use, as a trigger spray. A culture of the challenge microorganism was prepared in accordance with the EPA method (which references AOAC Method 960.09D). The inoculum suspension appeared appropriately turbid to produce 1 x 10^9 CFU/mL. The product was not tested in the presence of a 5% organic soil load. Polypropylene/Olefin carpet was cut into 8 inch x 12 inch carpet pieces, and six 2 inch x 2 inch square carriers were cut into the larger carpet pieces. The carpet was fastened to a mounting tray and sterilized by autoclave. Each cut square carrier was inoculated with 100 µL of a 22.75 hour old suspension of test organism. The inoculum was spread evenly over the surface of each cut square carrier. The cut square carriers were dried in an incubator for 60 minutes at 35-37°C with sterile foil loosely covering the top. For the single lot of product, separate cut square carriers were sprayed (16 pumps) with the product from a distance of 4-6 inches from the carrier surface. Each cut square carrier was then scrubbed for 30 seconds using 30 circular clockwise strokes and 30 circular counterclockwise strokes. A circular area of pile ~3 inches in diameter around the center of each cut square carrier was scrubbed. A new brush was used for each cut square carrier. The cut square carriers remained at 24.1°C for 60 minutes. Following exposure, each cut square carrier was removed from the larger carpet piece and transferred to a sterile vessel containing 100 mL of Letheen Broth with 0.07% Lecithin and 0.5% Tween 80 and 10 stainless steel penicylinders. Each vessel was shaken vigorously for 1 minute at 200 RPM to free the bacteria from the carpet fibers. Ten-fold serial dilutions were

prepared and 1.00 mL of the 10^0 through 10^{-3} dilutions were plated in duplicate on tryptic soy agar with 5% sheep's blood. All subcultures were incubated for 44.25 hours at 25-30°C. Following incubation, the subcultures were enumerated. Controls included those for unscrubbed population count, scrubbed population count, purity, sterility, and neutralization confirmation (the single product lot).

21. MRID 485735-21 "Carpet Sanitizer, Test Organism: *Pseudomonas aeruginosa* (ATCC 15442)" for Benefect Daily Disinfectant-Cleaner, by Becky Lien. Study conducted at ATS Labs. Study completion date – September 21, 2010. Project Number A09878.

This study was conducted against *Pseudomonas aeruginosa* (ATCC 15442). One lot (Lot No. FM040210001) of the product, Benefect Daily Disinfectant-Cleaner, was tested. The laboratory report referenced the Carpet Sanitizer Test from DIS/TSS-8. The product was received ready-to-use, as a trigger spray. A culture of the challenge microorganism was prepared in accordance with the EPA method (which references AOAC Method 960.09D). The inoculum suspension was compared to a 4.0 McFarland turbidity standard to produce an average of ~1.0×10^9 CFU/mL. The product was not tested in the presence of a 5% organic soil load. Nylon carpet was cut into 8 inch x 12 inch carpet pieces, and six 2 inch x 2 inch square carriers were cut into the larger carpet pieces. The carpet was fastened to a mounting tray and sterilized by autoclave. Each cut square carrier was inoculated with 100 µL of an 18-24 hour old suspension of test organism. The inoculum was spread evenly over the surface of each cut square carrier. The cut square carriers were dried in an incubator for 60 minutes at 35-37°C with sterile foil loosely covering the top. For the single lot of product, separate cut square carriers were sprayed (15 pumps) with the product from a distance of ~6 inches from the carrier surface until thoroughly wet. Each cut square carrier was then scrubbed for 30 seconds using 30 circular clockwise strokes and 30 circular counterclockwise strokes. A circular area of pile ~3 inches in diameter around the center of each cut square carrier was scrubbed. A new brush was used for each cut square carrier. The cut square carriers remained at 23.9°C for 60 minutes. Following exposure, each cut square carrier was removed from the larger carpet piece and transferred to a sterile vessel containing 100 mL of Letheen Broth with 0.07% Lecithin and 0.5% Tween 80 and 10 stainless steel penicylinders. Each vessel was shaken vigorously for 1 minute at 200 RPM to free the bacteria from the carpet fibers. Ten-fold serial dilutions were prepared and 1.00 mL of the 10^0 through 10^{-3} dilutions were plated in duplicate on tryptic soy agar with 5% sheep's blood. All subcultures were incubated for ~46.25 hours at 35-37°C. The subcultures were stored for 1 day at 2-8°C prior to examination. Following incubation and storage, the subcultures were enumerated. Controls included those for unscrubbed population count, scrubbed population count, purity, sterility, viability, and neutralization confirmation (the single product lot).

22. MRID 485735-22 "Carpet Sanitizer, Test Organism: *Pseudomonas aeruginosa* (ATCC 15442)" for Benefect Daily Disinfectant-Cleaner, by Becky Lien. Study conducted at ATS Labs. Study completion date – September 22, 2010. Project Number A09880.

This study was conducted against *Pseudomonas aeruginosa* (ATCC 15442). One lot (Lot No. D089662) of the product, Benefect Daily Disinfectant-Cleaner, was tested. The laboratory report referenced the Carpet Sanitizer Test from DIS/TSS-8. The product lot tested was at least 60 days old at the time of testing. The product was received ready-to-use, as a trigger spray. A culture of the challenge microorganism was prepared in accordance with the

EPA method (which references AOAC Method 960.09D). The inoculum suspension was compared to a 4.0 McFarland turbidity standard to produce an average of ~1.0 x 10^9 CFU/mL. The product was not tested in the presence of a 5% organic soil load. Nylon carpet was cut into 8 inch x 12 inch carpet pieces, and six 2 inch x 2 inch square carriers were cut into the larger carpet pieces. The carpet was fastened to a mounting tray and sterilized by autoclave. Each cut square carrier was inoculated with 100 µL of an 18-24 hour old suspension of test organism. The inoculum was spread evenly over the surface of each cut square carrier. The cut square carriers were dried in an incubator for 60 minutes at 35-37°C with sterile foil loosely covering the top. For the single lot of product, separate cut square carriers were sprayed (15 pumps) with the product from a distance of ~6 inches from the carrier surface until thoroughly wet. Each cut square carrier was then scrubbed for 30 seconds using 30 circular clockwise strokes and 30 circular counterclockwise strokes. A circular area of pile ~3 inches in diameter around the center of each cut square carrier was scrubbed. A new brush was used for each cut square carrier. The cut square carriers remained at 23.9°C for 60 minutes. Following exposure, each cut square carrier was removed from the larger carpet piece and transferred to a sterile vessel containing 100 mL of Letheen Broth with 0.07% Lecithin and 0.5% Tween 80 and 10 stainless steel penicylinders. Each vessel was shaken vigorously for 1 minute at 200 RPM to free the bacteria from the carpet fibers. Ten-fold serial dilutions were prepared and 1.00 mL of the 10^0 through 10^{-3} dilutions were plated in duplicate on tryptic soy agar with 5% sheep's blood. All subcultures were incubated for ~46.25 hours at 35-37°C. Following incubation, the subcultures were enumerated. Controls included those for unscrubbed population count, scrubbed population count, purity, sterility, viability, and neutralization confirmation (the single product lot).

23. MRID 485735-23 "Carpet Sanitizer, Test Organism: *Pseudomonas aeruginosa* (ATCC 15442)" for Benefect Daily Disinfectant-Cleaner, by Lynsey Wieland. Study conducted at ATS Labs. Study completion date – December 20, 2010. Project Number A10610.

This study was conducted against *Pseudomonas aeruginosa* (ATCC 15442). One lot (Lot No. FM100710001) of the product, Benefect Daily Disinfectant-Cleaner, was tested. The laboratory report referenced the Carpet Sanitizer Test from DIS/TSS-8. The product lot tested was at least 60 days old at the time of testing. The product was received ready-to-use, as a trigger spray. A culture of the challenge microorganism was prepared in accordance with the EPA method (which references AOAC Method 960.09D). The inoculum suspension was adjusted. The inoculum had a final spectrophotometer value (at 620 nm) of 1.973. The product was not tested in the presence of a 5% organic soil load. Nylon carpet was cut into 8 inch x 12 inch carpet pieces, and six 2 inch x 2 inch square carriers were cut into the larger carpet pieces. The carpet was fastened to a mounting tray and sterilized by autoclave. Each cut square carrier was inoculated with 100 µL of a 23.5 hour old suspension of test organism. The inoculum was spread evenly over the surface of each cut square carrier. The cut square carriers were dried in an incubator for 60 minutes at 35-37°C with sterile foil loosely covering the top. For the single lot of product, separate cut square carriers were sprayed (20 pumps) with the product from a distance of ~4-6 inches from the carrier surface. Each cut square carrier was then scrubbed for 30 seconds using 30 circular clockwise strokes and 30 circular counterclockwise strokes. A circular area of pile ~3 inches in diameter around the center of each cut square carrier was scrubbed. A new brush was used for each cut square carrier. The cut square carriers remained at 22.4°C for 60 minutes. Following exposure, each cut square carrier was removed from the larger carpet piece and transferred to a sterile vessel containing 100 mL of Letheen Broth with 0.07% Lecithin and 0.5% Tween 80 and 10 stainless steel penicylinders. Each vessel was shaken vigorously for 1 minute at ~200 RPM to free the bacteria from the carpet fibers. Ten-

fold serial dilutions were prepared and 1.00 mL of the 10^0 through 10^{-3} dilutions were plated in duplicate on agar. All subcultures were incubated for 45.25 hours at 35-37°C. Following incubation, the subcultures were enumerated. Controls included those for unscrubbed population count, scrubbed population count, purity, sterility, and neutralization confirmation (the single product lot).

24. MRID 485735-24 "Carpet Sanitizer, Test Organism: *Pseudomonas aeruginosa* (ATCC 15442)" for Benefect Daily Disinfectant-Cleaner, by Lynsey Wieland. Study conducted at ATS Labs. Study completion date – September 16, 2010. Project Number A09875.

This study was conducted against *Pseudomonas aeruginosa* (ATCC 15442). One lot (Lot No. FM040210001) of the product, Benefect Daily Disinfectant-Cleaner, was tested. The laboratory report referenced the Carpet Sanitizer Test from DIS/TSS-8. The product lot tested was at least 60 days old at the time of testing. The product was received ready-to-use, as a trigger spray. A culture of the challenge microorganism was prepared in accordance with the EPA method (which references AOAC Method 960.09D). The inoculum suspension was compared to a 4.0 McFarland turbidity standard to produce an average of ~1.0 x 10^9 CFU/mL. The product was not tested in the presence of a 5% organic soil load. Polypropylene/Olefin carpet was cut into 8 inch x 12 inch carpet pieces, and six 2 inch x 2 inch square carriers were cut into the larger carpet pieces. The carpet was fastened to a mounting tray and sterilized by autoclave. Each cut square carrier was inoculated with 100 µL of a ~22 hour old suspension of test organism. The inoculum was spread evenly over the surface of each cut square carrier. The cut square carriers were dried in an incubator for 60 minutes at 35-37°C with sterile foil loosely covering the top. For the single lot of product, separate cut square carriers were sprayed (15 pumps) with the product from a distance of ~6 inches from the carrier surface until thoroughly wet. Each cut square carrier was then scrubbed for 30 seconds using 30 circular clockwise strokes and 30 circular counterclockwise strokes. A circular area of pile ~3 inches in diameter around the center of each cut square carrier was scrubbed. A new brush was used for each cut square carrier. The cut square carriers remained at 23.9°C for 60 minutes. Following exposure, each cut square carrier was removed from the larger carpet piece and transferred to a sterile vessel containing 100 mL of Letheen Broth with 0.07% Lecithin and 0.5% Tween 80 and 10 stainless steel penicylinders. Each vessel was shaken vigorously for 1 minute at 200 RPM to free the bacteria from the carpet fibers. Ten-fold serial dilutions were prepared and 1.00 mL of the 10^0 through 10^{-3} dilutions were plated in duplicate on tryptic soy agar with 5% sheep's blood. All subcultures were incubated for ~46.25 hours at 35-37°C. Following incubation, the subcultures were enumerated. Controls included those for unscrubbed population count, scrubbed population count, purity, sterility, viability, and neutralization confirmation (the single product lot).

25. MRID 485735-25 "Carpet Sanitizer, Test Organism: *Pseudomonas aeruginosa* (ATCC 15442)" for Benefect Daily Disinfectant-Cleaner, by Lynsey Wieland. Study conducted at ATS Labs. Study completion date – September 16, 2010. Project Number A09876.

This study was conducted against *Pseudomonas aeruginosa* (ATCC 15442). One lot (Lot No. D119725) of the product, Benefect Daily Disinfectant-Cleaner, was tested. The laboratory report referenced the Carpet Sanitizer Test from DIS/TSS-8. The product lot tested

was at least 60 days old at the time of testing. The product was received ready-to-use, as a trigger spray. A culture of the challenge microorganism was prepared in accordance with the EPA method (which references AOAC Method 960.09D). The inoculum suspension was compared to a 4.0 McFarland turbidity standard to produce an average of ~1.0 x 10^9 CFU/mL. The product was not tested in the presence of a 5% organic soil load. Polypropylene/Olefin carpet was cut into 8 inch x 12 inch carpet pieces, and six 2 inch x 2 inch square carriers were cut into the larger carpet pieces. The carpet was fastened to a mounting tray and sterilized by autoclave. Each cut square carrier was inoculated with 100 µL of a ~22 hour old suspension of test organism. The inoculum was spread evenly over the surface of each cut square carrier. The cut square carriers were dried in an incubator for 60 minutes at 35-37°C with sterile foil loosely covering the top. For the single lot of product, separate cut square carriers were sprayed (15 pumps) with the product from a distance of ~6 inches from the carrier surface, until thoroughly wet. Each cut square carrier was then scrubbed for 30 seconds using 30 circular clockwise strokes and 30 circular counterclockwise strokes. A circular area of pile ~3 inches in diameter around the center of each cut square carrier was scrubbed. A new brush was used for each cut square carrier. The cut square carriers remained at 23.9°C for 60 minutes. Following exposure, each cut square carrier was removed from the larger carpet piece and transferred to a sterile vessel containing 100 mL of Letheen Broth with 0.07% Lecithin and 0.5% Tween 80 and 10 stainless steel penicylinders. Each vessel was shaken vigorously for 1 minute at 200 RPM to free the bacteria from the carpet fibers. Ten-fold serial dilutions were prepared and 1.00 mL of the 10^0 through 10^{-3} dilutions were plated in duplicate on tryptic soy agar with 5% sheep's blood. All subcultures were incubated for ~46.25 hours at 35-37°C. The subcultures were stored for 1 day at 2-8°C prior to examination. Following incubation and storage, the subcultures were enumerated. Controls included those for unscrubbed population count, scrubbed population count, purity, sterility, viability, and neutralization confirmation (the single product lot).

26. MRID 485735-26 "Carpet Sanitizer, Test Organism: *Pseudomonas aeruginosa* (ATCC 15442)" for Benefect Daily Disinfectant-Cleaner, by Lynsey Wieland. Study conducted at ATS Labs. Study completion date – December 28, 2010. Project Number A10609.

This study was conducted against *Pseudomonas aeruginosa* (ATCC 15442). One lot (Lot No. FM100710001) of the product, Benefect Daily Disinfectant-Cleaner, was tested. The laboratory report referenced the Carpet Sanitizer Test from DIS/TSS-8. The product lot tested was at least 60 days old at the time of testing. The product was received ready-to-use, as a trigger spray. A culture of the challenge microorganism was prepared in accordance with the EPA method (which references AOAC Method 960.09D. The inoculum suspension had a final spectrophotometer value (at 620 nm) of 1.973. The product was not tested in the presence of a 5% organic soil load. Polyproplyene/Olefin carpet was cut into 8 inch x 12 inch carpet pieces, and six 2 inch x 2 inch square carriers were cut into the larger carpet pieces. The carpet was fastened to a mounting tray and sterilized by autoclave. Each cut square carrier was inoculated with 100 µL of a 23.5 hour old suspension of test organism. The inoculum was spread evenly over the surface of each cut square carrier. The cut square carriers were dried in an incubator for 60 minutes at 35-37°C with sterile foil loosely covering the top. For the single lot of product, separate cut square carriers were sprayed (20 pumps) with the product from a distance of ~4-6 inches from the carrier surface. Each cut square carrier was then scrubbed for 30 seconds using 30 circular clockwise strokes and 30 circular counterclockwise strokes. A circular area of pile ~3 inches in diameter around the center of each cut square carrier was scrubbed. A new brush was used for each cut square carrier. The cut square carriers remained at 24.3°C for 60

minutes. Following exposure, each cut square carrier was removed from the larger carpet piece and transferred to a sterile vessel containing 100 mL of Letheen Broth with 0.07% Lecithin and 0.5% Tween 80 and 10 stainless steel penicylinders. Each vessel was shaken vigorously for 1 minute at ~200 RPM to free the bacteria from the carpet fibers. Ten-fold serial dilutions were prepared and 1.00 mL of the 10^0 through 10^{-3} dilutions were plated in duplicate on tryptic soy agar with 5% sheep's blood. All subcultures were incubated for 45.25 hours at 35-37°C. Following incubation, the subcultures were enumerated. Controls included those for unscrubbed population count, scrubbed population count, purity, sterility, and neutralization confirmation (the single product lot).

V RESULTS

MRID Number	Organism	No. Exhibiting Growth/ Total No. Tested		Carrier Population (CFU/ carrier)
		Lot No. FM093009003	Lot No. FM093009004	
10-Minute Exposure Time				
485735-02	*Escherichia coli* O157:H7	0/10	0/10	1.31×10^5
485735-03	Community Acquired Methicillin Resistant *Staphylococcus aureus* (NARSA NRS384)	0/10	---	1.38×10^6
485735-04	Community Acquired Methicillin Resistant *Staphylococcus aureus* (NARSA NRS384)	---	0/10	1.61×10^6
		Lot No. D089662	**Lot No. D119725**	
485735-05	*Trichophyton mentagrophytes*	0/10	---	1.1×10^4
485735-06	*Trichophyton mentagrophytes*	---	0/10	1.2×10^4

MRID Number	Organism	Results			Dried Virus Count
			LC# 071J3012	LC# 071F1302	
10-Minute Exposure Time					
485735-01	Avian influenza A (H3N2) virus	10^{-1} to 10^{-7} dilutions	Complete inactivation	Complete inactivation	$10^{4.5}$ $TCID_{50}$/0.1 mL
		$TCID_{50}$/0.1 mL	$\leq 10^{0.5}$	$\leq 10^{0.5}$	

MRID Number	Organism	No. Exhibiting Growth/ Total No. Tested		Control Tiles
		Lot No. FM100710001	Lot No. FM100710002	
485735-07	*Aspergillus niger*	0/10	---	10/10*
485735-08	*Aspergillus niger*	---	0/10	10/10*

* At least 50% fungal growth on each untreated control tile was observed.

MRID Number	Organism	Lot No.	Total No. Surviving	Scrubbed Population Count	Percent Reduction
			(CFU/carrier)		
60-Minute Exposure Time; Nylon Carpet					
485735-09	*Staphylococcus aureus*	FM040210001	$<3 \times 10^2$	4.9×10^7	>99.9
485735-10	*Staphylococcus aureus*	D089662	7.8×10^3	1.33×10^8	>99.9
485735-11	*Staphylococcus aureus*	D119725	1.3×10^4	5.7×10^7	>99.9
485735-15	*Enterobacter aerogenes*	FM040210001	3.1×10^4	8.5×10^7	>99.9
485735-16	*Enterobacter aerogenes*	FM100710001	1.0×10^4	1.6×10^7	>99.9
485735-17	*Enterobacter aerogenes*	FM100710002	$<1.0 \times 10^2$	9.12×10^6	>99.9†
485735-21	*Pseudomonas aeruginosa*	FM040210001	2.82×10^4	9.12×10^7	>99.9
485735-22	*Pseudomonas aeruginosa*	D089662	7.08×10^4	9.12×10^7	>99.9
485735-23	*Pseudomonas aeruginosa*	FM100710001	1.00×10^4	1.6×10^7	>99.9
60-Minute Exposure Time; Polypropylene/Olefin Carpet					
485735-12	*Staphylococcus aureus*	FM100710001	$<1.0 \times 10^2$	1.82×10^7	>99.9
485735-13	*Staphylococcus aureus*	FM100710002	$<6.3 \times 10^2$	9.12×10^7	>99.9
485735-14	*Staphylococcus aureus*	FM040210001	$<1.6 \times 10^2$	1.10×10^7	>99.9
485735-18	*Enterobacter aerogenes*	FM100710001	$<4.0 \times 10^2$	1.45×10^7	>99.9
485735-19	*Enterobacter aerogenes*	FM100710002	$<1.3 \times 10^2$	7.08×10^6	>99.9
485735-20	*Enterobacter aerogenes*	FM040210001	$<2.0 \times 10^2$	1.26×10^6	>99.9
485735-24	*Pseudomonas aeruginosa*	FM040210001	3.98×10^4	4.436×10^8	>99.9
485735-25	*Pseudomonas aeruginosa*	D119725	3.55×10^5	4.436×10^8	>99.9
485735-26	*Pseudomonas aeruginosa*	FM100710001	2.5×10^2	6.368×10^6	>99.9

†Note: The unscrubbed population control result did not meet the acceptance criterion. The Study Director did not believe that this deviation impacted the validity of the study as the amount of organism recovered on the scrubbed population control exceeded the acceptance criterion for the unscrubbed population control.

VI CONCLUSIONS

1. The submitted efficacy data support the use of the product, CleanWell Daily Disinfecting Cleaner and CleanWell Daily Disinfectant Cleaner (i.e., Benefect Botanical Daily Cleaner Disinfectant Spray), as a disinfectant with bactericidal activity against the following microorganisms on hard, non-porous surfaces in the presence of a 5% organic soil load for a 10-minute contact time:

Escherichia coli O157:H7	MRID 485735-02
Community Acquired Methicillin Resistant *Staphylococcus aureus* (NARSA NRS384)	MRID 485735-03 and -04

Complete killing was observed in the subcultures of the required number of carriers tested against the required number of product lots. Neutralization confirmation testing showed positive growth of the microorganisms. Viability controls were positive for growth. Purity controls were reported as pure. Sterility controls did not show growth.

2. The submitted efficacy data (MRID 485735-05 and -06) support the use of the product, Benefect Daily Disinfectant Cleaner (i.e., Benefect Botanical Daily Cleaner Disinfectant Spray), as a disinfectant with fungicidal activity against *Trichophyton mentagrophytes* on hard, non-porous surfaces in the presence of a 5% organic soil load for a 10-minute contact time. Complete killing was observed in the subcultures of the required number of carriers tested against the required number of product lots. Neutralization confirmation testing showed positive growth of the microorganism. Viability controls were positive for growth. Purity controls were reported as pure. Sterility controls did not show growth.

3. The submitted efficacy data (MRID 485735-01) support the use of the product, Benefect Botanical Daily Cleaner Disinfectant (i.e., Benefect Botanical Daily Cleaner Disinfectant Spray), as a disinfectant with virucidal activity against Avian influenza A (H3N2) virus on hard, non-porous surfaces in the presence of a 5% organic soil load for a 10-minute contact time. A recoverable virus titer of at least 10^4 was achieved. Cytotoxicity was not observed. Complete inactivation (no growth) was indicated in all dilutions tested.

4. The submitted efficacy data (MRID 485735-07 and -08) support the use of the product, Benefect Daily Disinfectant Cleaner (i.e., Benefect Botanical Daily Cleaner Disinfectant Spray), as a mildewstat against *Aspergillus niger* on pre-cleaned, hard, non-porous surfaces. No growth was observed 7 days after treatment. Testing was conducted on 2 product lots. Untreated control tiles exhibited growth of *Aspergillus niger* on 50% to 98% of each untreated tile surface. Purity controls were reported as pure. Sterility controls did not show growth.

5. The submitted efficacy data support the use of the product, Benefect Daily Disinfectant-Cleaner (i.e., Benefect Botanical Daily Cleaner Disinfectant Spray), as a carpet sanitizer against the following microorganisms on pre-cleaned carpet surfaces for a 60-minute contact time:

<u>Nylon Carpet</u>	
Staphylococcus aureus	MRID 485735-09, -10, and -11
Enterobacter aerogenes	MRID 485735-15, -16, and -17
Pseudomonas aeruginosa	MRID 485735-21, -22, and -23
<u>Polypropylene/Olefin Carpet</u>	
Staphylococcus aureus	MRID 485735-12, -13, and -14
Enterobacter aerogenes	MRID 485735-18, -19, and -20
Pseudomonas aeruginosa	MRID 485735-24, -25, and -26

Bacterial reductions of at least 99.9 percent over the scrubbed population control were observed within 60 minutes. In testing against *Staphylococcus aureus* and *Enterobacter aerogenes*, at least one of the product lots tested was at least 60 days old at the time of testing. Neutralization confirmation testing met the acceptance criterion of growth within 1 $\log_{10}$ of the corresponding neutralization confirmation population control. When conducted, viability controls were positive for growth. Purity controls were reported as pure. Sterility controls did not show growth, with one exception. Growth of Gram positive bacilli was noted for one carrier sterility control during testing against *Enterobacter aerogenes* (see MRID 485735-15).

VII RECOMMENDATIONS

1. The proposed label claims that the product, Benefect Botanical Daily Cleaner Disinfectant Spray, is an effective disinfectant against the following microorganisms on hard, non-porous surfaces in the presence of light to moderate soil loads for a 10-minute contact time:

Community Acquired Methicillin Resistant *Staphylococcus aureus*
Escherichia coli O157:H7
Methicillin Resistant *Staphylococcus aureus* (NARSA NRS384)

Trichophyton mentagrophytes
Influenza A (H3N2) virus

These claims are acceptable as they are supported by the submitted data.

2. The proposed label claims that the product, Benefect Botanical Daily Cleaner Disinfectant Spray, kills and prevents mold and mildew for 7 days, specifically *Aspergillus niger*. This claim is acceptable as it is supported by the submitted data; however, directions for use of the product as a mildewstat must be added to the proposed label.

3. The proposed label claims that the product, Benefect Botanical Daily Cleaner Disinfectant Spray, is effective in sanitizing pre-cleaned nylon and olefin carpets against *Staphylococcus aureus*, *Enterobacter aerogenes*, and *Pseudomonas aeruginosa* for a 60-minute contact time. This claim is acceptable as it is supported by the submitted data.

4. Human immunodeficiency virus is identified on the proposed label and Data Matrix (i.e., MRID 479498-10); however, it was not identified on the last accepted label. The letter from the applicant's representative to EPA (dated August 16, 2011) suggests that efficacy studies were provided to support the product, Benefect Botanical Daily Cleaner Disinfectant Towelette. The data did demonstrate efficacy against the spray at the ready to use preparation for 10 minutes in the presence of 5% organic soil load.

5. *Candida albicans* is identified on the proposed label and Data Matrix (i.e., MRID 479498-11 and 479498-12); however, it is not identified on the last accepted label. The letter from the applicant's representative to EPA (dated August 16, 2011) suggests that efficacy studies were provided to support the product, Benefect Botanical Daily Cleaner Disinfectant Towelette. The data did demonstrate efficacy against the spray at the ready to use preparation for 10 minutes in the presence of 5% organic soil load.

6. The following revisions to the proposed label are recommended:

- On page 3 of the proposed label, change "Scrub or agitate the carpet as need" to read "Scrub or agitate the carpet as needed."
- On page 3 of the proposed label, change "test substance" to read "product."
- On page 6 of the proposed label, general cold claims are require testing against Rhinovirus, Coronavirus and RSV.

- On page 6 of the proposed label, the claim "bacteriostat" and "bacteriostatic" are unacceptable. The disinfectant assessment is –cidal, and claims should be limited to that claim.

- On page 6 of the proposed label, the claim "power" is unacceptable as implies heighten efficacy.

- On page 6 of the proposed label, the claim "[10 minute] sanitizer" is unacceptable.

- On page 8 of the proposed label, remove the reference option to Influenza A virus (ATCC VR-1469), as efficacy data was not generated to support this claim.

- On pages 10 and 18 of the proposed label, remove the term "botanically derived".

- On page 14 of the proposed label, change "fiberglass" to read "sealed fiberglass." Fiberglass is a porous surface.

- On page 14 of the proposed label, change "marble" to read "sealed marble." Marble is a porous surface.

- On page 14 of the proposed label, change "stone" to read "sealed stone." Stone is a porous surface.

- On page 15 of the proposed label, remove the statement "Only disinfectant with this amazing low-streak cleaning formula".

- On page 22 of the proposed label, remove the statement "Contains no endocrine disruptors".

- On page 25 of the proposed label, change "Methicillin Resistant *S. aureus* (MRSA)" to read "Community Acquired Methicillin Resistant *Staphylococcus aureus* (CA-MRSA)."

- On page 25 of the proposed label, identify the ATCC number for *Escherichia coli* as "ATCC 11229."

- On page 25 of the proposed label, add the following bacterium and its ATCC number: *Escherichia coli* O157:H7 (ATCC 35150).

- On the proposed label, quantitative assessments (i.e. Kills over 99.9[9]% are reserved for quantitative tests. The Germicidal Spray Test is a qualitative assessment.

- When quantitative assessments are proposed on the label, they must be qualified to the actual test organism and level of efficacy (food contact sanitizer, sanitizer, etc.).

7. The following revision to the Data Matrix is recommended:

   - Include a subheading for studies supporting hard, non-porous surface sanitizing claims, such as "Hard Surface Sanitizer."